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Homology of the 70-kilodalton antigens from Mycobacterium leprae and Mycobacterium bovis with the Mycobacterium tuberculosis 71-kilodalton antigen and with the conserved heat shock protein 70 of eucaryotes.

Garsia RJ, Hellqvist L, Booth RJ, Radford AJ, Britton WJ, Astbury L, Trent RJ, Basten A.

Clinical Immunology Research Centre, University of Sydney, New South Wales, Australia.

Two lambda gt11 recombinant clones, JKL2 and JKL15, each containing an insert coding for part of the highly immunogenic 70-kilodalton (kDa) protein antigen, were isolated from a Mycobacterium leprae genomic library by immunoscreening with the monoclonal antibody L7. Clone JKL2 contained the largest insert, 2.3 kilobase pairs. Nonoverlapping fragments of this insert were used as probes and showed strong hybridization to a number of Mycobacterium tuberculosis-lambda gt11 recombinants producing proteins recognized by an anti-M. tuberculosis 71-kDa monoclonal antibody, IT11. One clone from a recombinant Mycobacterium bovis library was also characterized by using L7, and the insert from this clone, B5bt, hybridized strongly to the M. leprae probes as well. The nucleotide sequence of the 1,037-base-pair coding region of the JKL2 M. leprae clone which encodes the carboxy-terminal half of the 70-kDa protein had extensive homology with genes from a number of species. In all cases, these genes, including the recently described Ag63 and Ag361 of Plasmodium falciparum, were found to be members of the heat shock protein 70 (hsp 70) family of genes. At the amino acid level, homology was maximal between amino acids 83 through 107 and 159 through 184, which showed extreme conservation (92 and 85% identity) with Escherichia coli DnaK amino acids 386 through 409 and 460 through 485, respectively, and was 51% homologous over the entire coding region (amino acids 1 through 344 of JKL2). In contrast, amino acids 129 through 158 had maximal homology with the phylogenetically more distant Xenopus laevis hsp70. Homology declined substantially in the carboxy-terminal 34 amino acids. The predicted ATP-binding functional activity of the 70-kDa antigen from M. bovis was confirmed with affinity purification of the antigen by binding to ATP-agarose and elution with ATP. In view of the conservation of sequences between these mycobacterial antigens and mammalian endogenous cellular enzymes, further evaluation of these molecules in vivo may aid in understanding tolerance to self-antigens as well as provide potentially useful

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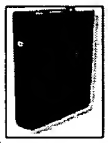

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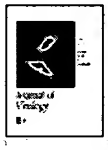

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Annu. Rev. Immunol. 2002. 20:395-425.

INTERACTION OF HEAT SHOCK PROTEINS WITH PEPTIDES AND ANTIGEN PRESENTING CELLS: Chaperoning of the Innate and Adaptive Immune Responses

Pramod Srivastava
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KEY WORDS: dendritic cells, cross-priming, indirect presentation, cancer, infectious diseases

Heat shock proteins are abundant soluble intracellular proteins, present in all cells. Members of the heat shock protein family bind peptides including antigenic peptides generated within cells. Heat shock proteins also interact with antigen presenting cells through CD91 and other receptors, eliciting a cascade of events including re-presentation of heat shock protein-chaperoned peptides by MHC, translocation of NFκB into the nuclei and maturation of dendritic cells. These consequences point to a key role of heat shock proteins in fundamental immunological phenomena such as activation of antigen presenting cells, indirect presentation (or cross-priming), and chaperoning of peptides during antigen presentation. Heat shock proteins appear to have been involved in innate immune responses since the emergence of phagocytes in early multicellular organisms and to have been commandeered for adaptive immune responses with the advent of specificity. These properties of heat shock proteins also allow them to be used for immunotherapy of cancers and infections in novel ways.

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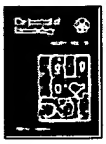

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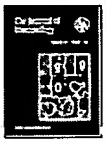

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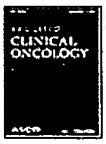

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Immunity, Vol. 12, 263-272, March, 2000

A Proposed Mechanism for the Induction of Cytotoxic T Lymphocyte Production by Heat Shock Fusion Proteins

Bryan K. Cho [1], Deborah Palliser [1], Eduardo Guillen [3], Jan Wisniewski [3], Richard A. Young [1,2], Jianzhu Chen [1], and Herman N. Eisen [1]

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A 65 kDa mycobacterial heat shock protein (hsp65), fused to a polypeptide that contains an octapeptide (SIYRYYGL) agonist for a particular T cell receptor (2C TCR), stimulated C57BL/6 mice as well as CD4-deficient mice to produce $CD8^+$ cytolytic T lymphocytes (CTL) to the fusion partner's octapeptide. This and other hsp65 fusion proteins but not native hsp65 itself stimulated dendritic cells in vitro and in vivo to upregulate the levels of MHC (class I and II) and costimulatory (B7.2) molecules. The results suggest a mechanism for the general finding that hsp fusion proteins, having fusion partners of widely differing lengths and sequences, elicit CD8 CTL to peptides from the fusion partners without requiring exogenous adjuvants or the participation of $CD4^+$ T cells.

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